ABSTRACT OF THE DISCLOSURE

We describe a new two-step culture method for mass production *in vitro* of erythroid cells from either $CD34^+$ (10^5 cells/mL) or light-density (10^6 cells/mL) cells purified from the blood of normal donors and thalassemic patients. The method includes (i) culture of the cells in the presence of dexamethasone and estradiol (10^{-6} M each) and (ii) the growth factors SCF (50 ng/mL), IL-3 (1 ng/mL), and EPO (1 U/mL). In their proliferative phase, these cultures generated about 1–2 x 10^7 erythroblasts for each milliliter of blood collected from normal donors or thalassemic patients. They were composed mostly (90%) of $CD45^{low}$/glycophorin $(GPA)^{neg}$ / $CD71^{low}$ cells at day 7, 50–60% of which became $CD45^{neg}$/GPA+/$CD71^{high}$ by days 15–20. However, when cells from days 7 to 12 of the proliferative phase were transferred in differentiation medium containing EPO and insulin, they progressed to mature erythroblasts (>90% $benzidine^{pos}$ and $CD45^{neg}$/GPA^+/$CD71^{medium}$) in 4 days. Because of the high number of erythroid cells that are generated from modest volumes of blood, this method will prove useful in donor-specific studies of erythroid differentiation.

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